WEST

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Main Menu | Search Form | Posting Counts | Show S Numbers | Edit S Numbers | Preferences | Cases

Search Results -

Terms	Documents
L5 and endotherm	2

Database: US Patents Full-Text Database; US Pre-Grant Publication Full-Text Database; JPO Abstracts Database; EPO Abstracts Database; Derwent World Patents Index; IBM Technical Disclosure Bulletins

Search: L6

Refine Search | Recall Text | Clear

Search History

DATE: Friday, August 02, 2002 Printable Copy Create Case

Set Name side by side	Query	Hit Count	Set Name result set
DB=USPT,PGPB,EPAB,DWPI; PLUR=YES; OP=OR			
L6	L5 and endotherm	2	L6
L5	L4 and viscosity	115	L5
L4	L3 and (increase$2 with amylose)	165	L4
L3	L2 and potato	2741	L3
L2	L1 and starch	5785	L2
L1	amylose	7206	L1

END OF SEARCH HISTORY

WEST

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Main Menu | Search Form | Posting Counts | Show S Numbers | Edit S Numbers | Preferences | Cases

Search Results -

Terms	Documents
L2 and (cauliflower with mosaic with virus)	3

Database: US Patents Full-Text Database; US Pre-Grant Publication Full-Text Database; JPO Abstracts Database; EPO Abstracts Database; Derwent World Patents Index; IBM Technical Disclosure Bulletins

Search: L3

Refine Search

Recall Text | Clear

Search History

DATE: Friday, August 02, 2002 Printable Copy Create Case

Set Name side by side	Query	Hit Count	Set Name result set
DB=USPT,PGPB,EPAB,DWPI; PLUR=YES; OP=OR			
L3	L2 and (cauliflower with mosaic with virus)	3	L3
L2	L1 and ("starch synthase III" or SSIII)	8	L2
L1	"starch synthase II" or SSII	75	L1

END OF SEARCH HISTORY

(FILE 'HOME' ENTERED AT 09:32:06 ON 02 AUG 2002)

FILE 'CAPLUS, BABS, CBNB, CEN, CIN, DKILIT, IFIPAT, JICST-EPLUS, PASCAL, PLASNEWS, PROMT, RAPRA, SCISEARCH, TEXTILETECH, USPATFULL, USPAT2, WPIDS, WTEXTILES' ENTERED AT 09:35:39 ON 02 AUG 2002

L1 29 S VISCOSITY ONSET
L2 16 S L1 AND STARCH
L3 12 S L2 AND POTATO

(FILE 'HOME' ENTERED AT 09:32:06 ON 02 AUG 2002)

FILE 'CAPLUS, BABS, CBNB, CEN, CIN, DKILIT, IFIPAT, JICST-EPLUS, PASCAL, PLASNEWS, PROMT, RAPRA, SCISEARCH, TEXTILETECH, USPATFULL, USPAT2, WPIDS, WTEXTILES' ENTERED AT 09:35:39 ON 02 AUG 2002

L1 29 S VISCOSITY ONSET
L2 16 S L1 AND STARCH
L3 12 S L2 AND POTATO

L3 ANSWER 1 OF 12 CAPLUS COPYRIGHT 2002 ACS
TI Improvements in or relating to plants and plant **starch** products resulting from transformation with antisense **starch** synthase constructs
AN 1999:811367 CAPLUS
DN 132:31779
TI Improvements in or relating to plants and plant **starch** products resulting from transformation with antisense **starch** synthase constructs
IN Edwards, Elizabeth Anne; Jobling, Stephen Alan; Martin, Catherine Rosemary; Schwall, Gerhard Peter; Smith, Alison Mary; Westcott, Roger John
PA National Starch and Chemical Investment Holding Corporation, USA
SO PCT Int. Appl., 50 pp.
CODEN: PIXXD2
DT Patent
LA English
IC ICM C12N015-54
ICS C12N015-82; C08B030-04; A01H005-00
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 9966050	A1	19991223	WO 1999-GB1902	19990615
	W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
	RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
	CA 2331300	AA	19991223	CA 1999-2331300	19990615
	AU 9943802	A1	20000105	AU 1999-43802	19990615
	EP 1092033	A1	20010418	EP 1999-926617	19990615
	R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
	JP 2002518015	T2	20020625	JP 2000-554859	19990615
PRAI	EP 1998-304716	A	19980615		
	WO 1999-GB1902	W	19990615		

RE.CNT 11 THERE ARE 11 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT
TI Improvements in or relating to plants and plant **starch** products resulting from transformation with antisense **starch** synthase constructs
AB A method for modifying plants by manipulating the activity of a combination of plant enzymes having **starch** synthase activity, in particular **starch** synthase II (SSII) and **starch** synthase III (SSIII). Modified plants, their use as food products and **starch**, in particular obtained from a modified **potato** plant, having novel properties and uses thereof are also disclosed. **Starch** extd. from **potato** plants transformed by introduction of and SSII/SSIII combination operably linked in the antisense orientation to a suitable promoter, has a **viscosity onset** temp. as detd. by viscoamylograph, which is significantly reduced compared to the effects predicted by reducing the 2 isoforms individually or in unmodified plants. The modified **starch** may have uses in food processing and other applications, such as in the paper, textiles, and adhesives industries (no data).
ST plant transformation antisense **starch** synthase; **potato** transformation antisense **starch** synthase
IT Adhesives
Barley

Cassava (Manihot esculenta)
Corn
Food processing
Paper
Pea
Plant (Embryophyta)
Potato (Solanum tuberosum)
Rice (Oryza sativa)
Textiles
Tomato
Transformation, genetic
Wheat
(improvements in or relating to plants and plant **starch** products resulting from transformation with antisense **starch** synthase constructs)

IT Antisense DNA
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(improvements in or relating to plants and plant **starch** products resulting from transformation with antisense **starch** synthase constructs)

IT Plasmid vectors
(pPOT17 and pSJ42 and pSJ119; improvements in or relating to plants and plant **starch** products resulting from transformation with antisense **starch** synthase constructs)

IT 9030-10-8, **Starch** synthase
RL: BPR (Biological process); BSU (Biological study, unclassified); BUU (Biological use, unclassified); BIOL (Biological study); PROC (Process); USES (Uses)
(II and III; improvements in or relating to plants and plant **starch** products resulting from transformation with antisense **starch** synthase constructs)

IT 9005-25-8DP, **Starch**, modified, biological studies
RL: BMF (Bioindustrial manufacture); FFD (Food or feed use); NUU (Other use, unclassified); PRP (Properties); BIOL (Biological study); PREP (Preparation); USES (Uses)
(improvements in or relating to plants and plant **starch** products resulting from transformation with antisense **starch** synthase constructs)

L3 ANSWER 2 OF 12 CAPLUS COPYRIGHT 2002 ACS
TI Consequences of antisense RNA inhibition of **starch** branching enzyme activity on properties of **potato starch**
AN 1998:508745 CAPLUS
DN 129:214130
TI Consequences of antisense RNA inhibition of **starch** branching enzyme activity on properties of **potato starch**
AU Safford, Richard; Jobling, Steve A.; Sidebottom, Chris M.; Westcott, Roger J.; Cooke, David; Tober, Karen J.; Strongitharm, Barbara H.; Russell, Alison L.; Gidley, Michael J.
CS Biosciences Division, Unilever Research, Sharnbrook, MK 441LQ, UK
SO Carbohydrate Polymers (1998), 35(3-4), 155-168
CODEN: CAPOD8; ISSN: 0144-8617
PB Elsevier Science Ltd.
DT Journal
LA English
TI Consequences of antisense RNA inhibition of **starch** branching enzyme activity on properties of **potato starch**
AB Antisense constructs contg. cDNAs for **potato starch** branching enzyme (SBE) were introduced into **potato** (Solanum tuberosum L.). A population of transgenic plants were generated in which tuber SBE activity was reduced by between 5 and 98% of control values. No

significant differences in amylose content or amylopectin branch length profiles of transgenic tuber starches were obsd. as a function of tuber SBE activity. Starches obtained from low SBE activity plants showed elevated phosphorus content. 31P-NMR anal. showed that this was due to proportionate increases in both 3- and 6-linked **starch** phosphates. A consistent alteration in **starch** gelatinization properties was only obsd. when the level of SBE activity was reduced to below .apprx.5% of that of control values. Starches from these low SBE activity plants showed increases of up to 5.degree.C in d.s.c. peak temp. and **viscosity onset** temp. Studies on melting of crystallites obtained from linear (1 .fwdarw. 4)-.alpha.-D-glucan oligomers suggest that an av. difference of double helix length of about one glucose residue might be sufficient to account for the obsd. differences in gelatinization properties. It is postulated that the modification of gelatinization properties at low SBE activities is due to a subtle alteration in amylopectin branch patterns resulting in small changes in double helix lengths within granules.

ST **potato starch** property branching enzyme antisense

IT **Potato** (Solanum tuberosum)
(transgenic; consequences of antisense RNA inhibition of **starch** branching enzyme activity on properties of **potato starch**)

IT 7723-14-0, Phosphorus, biological studies 9037-22-3, Amylopectin
RL: BOC (Biological occurrence); BSU (Biological study, unclassified); BIOL (Biological study); OCCU (Occurrence)
(consequences of antisense RNA inhibition of **starch** branching enzyme activity on properties of **potato starch**)

IT 9005-25-8, **Starch**, biological studies
RL: BOC (Biological occurrence); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); OCCU (Occurrence)
(consequences of antisense RNA inhibition of **starch** branching enzyme activity on properties of **potato starch**)

IT 9001-97-2, **Starch** branching enzyme
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(consequences of antisense RNA inhibition of **starch** branching enzyme activity on properties of **potato starch**)

L3 ANSWER 3 OF 12 CAPLUS COPYRIGHT 2002 ACS

TI Cloning and expression of soluble **starch** synthase of **potato** tubers and use of the enzyme for producing modified **starch**

AN 1997:592269 CAPLUS

DN 127:187509

TI Cloning and expression of soluble **starch** synthase of **potato** tubers and use of the enzyme for producing modified **starch**

IN Smith, Alison Mary; Marshall, Jacqueline; Edwards, Elizabeth Ann; Martin, Catherine Rosemary

PA National Starch and Chemical Investment Holding Corporation, USA

SO Eur. Pat. Appl., 39 pp.
CODEN: EPXXDW

DT Patent

LA English

IC ICM C12N015-82
ICS C12N015-54; C12N009-10; C08B030-00; C12N005-10; A01H005-00

FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI EP 779363	A2	19970618	EP 1996-309004	19961211
EP 779363	A3	19980520		

R: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE

AU 9674268 A1 19970703 AU 1996-74268 19961211
AU 723475 B2 20000824
PRAI GB 1995-25353 A 19951212
TI Cloning and expression of soluble **starch** synthase of **potato** tubers and use of the enzyme for producing modified **starch**
AB The cDNA encoding a sol. **starch** synthase was isolated from **potato** (Solanum tuberosa cultivar Desiree) tubers and its amino acid sequence deduced. The purified enzyme exhibits 100-140 kDa on SDS-PAGE. A transgenic **potato** plant expressing the antisense sequence of sol. **starch** synthase produced **starch** having a **viscosity onset** temp., as detd. by differential scanning calorimetry, lowered by at .gtoreq.5.degree.C compared to **starch** extd. from the non-transformed plants. Reduced sol. **starch** synthase activity affected the shape of **starch** granules, but had little effects on the amylose content.
ST **potato** sol **starch** synthase cDNA sequence; transgenic plant **starch** modification
IT Breeding, plant
Molecular cloning
(cloning and expression of sol. **starch** synthase of **potato** tubers and use of enzyme for producing modified **starch**)
IT Gene, plant
RL: BOC (Biological occurrence); BSU (Biological study, unclassified); BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); OCCU (Occurrence); USES (Uses)
(cloning and expression of sol. **starch** synthase of **potato** tubers and use of enzyme for producing modified **starch**)
IT cDNA sequences
(for **potato** sol. **starch** synthase; cloning and expression of sol. **starch** synthase of **potato** tubers and use of enzyme for producing modified **starch**)
IT Protein sequences
(of **potato** sol. **starch** synthase; cloning and expression of sol. **starch** synthase of **potato** tubers and use of enzyme for producing modified **starch**)
IT Antisense DNA
RL: BUU (Biological use, unclassified); SPN (Synthetic preparation); BIOL (Biological study); PREP (Preparation); USES (Uses)
(of sol. **starch** synthase cDNA; cloning and expression of sol. **starch** synthase of **potato** tubers and use of enzyme for producing modified **starch**)
IT Viscosity
(of **starch**; cloning and expression of sol. **starch** synthase of **potato** tubers and use of enzyme for producing modified **starch**)
IT Barley
Cassava (Manihot esculenta)
Corn
Oat
Plant (Embryophyta)
Potato (Solanum tuberosum)
Rice (Oryza sativa)
Sweet **potato**
Tomato
Wheat
(transgenic plant producing modified **starch**; cloning and expression of sol. **starch** synthase of **potato** tubers and use of enzyme for producing modified **starch**)
IT 179734-85-1

RL: AGR (Agricultural use); BAC (Biological activity or effector, except adverse); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); USES (Uses)
(amino acid sequence of sol. **starch** synthase; cloning and expression of sol. **starch** synthase of **potato** tubers and use of enzyme for producing modified **starch**)

IT 9005-25-8P, **Starch**, preparation
RL: BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
(cloning and expression of sol. **starch** synthase of **potato** tubers and use of enzyme for producing modified **starch**)

IT 173758-43-5
RL: AGR (Agricultural use); BOC (Biological occurrence); BSU (Biological study, unclassified); BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); OCCU (Occurrence); USES (Uses)
(nucleotide sequence for sol. **starch** synthase; cloning and expression of sol. **starch** synthase of **potato** tubers and use of enzyme for producing modified **starch**)

IT 9030-10-8, **Starch** synthase
RL: BAC (Biological activity or effector, except adverse); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(sol.; cloning and expression of sol. **starch** synthase of **potato** tubers and use of enzyme for producing modified **starch**)

L3 ANSWER 4 OF 12 IFIPAT COPYRIGHT 2002 IFI

have it

TI HIGH AMYLOSE **STARCH** FROM TRANSGENIC **POTATO** PLANTS; EXTRACTING **STARCH** FROM TUBERS WHICH HAVE **STARCH** BRANCHING ENZYME CDNA SEQUENCE OPERABLY LINKED IN ANTISENSE ORIENTATION TO PROMOTER; HAS ELEVATED PEAK TEMPERATURE OF GELATINIZATION AND ELEVATED VICOSITY ONSET TEMPERATURE

AN 3368895 IFIPAT;IFIUDB;IFICDB

TI HIGH AMYLOSE **STARCH** FROM TRANSGENIC **POTATO** PLANTS; EXTRACTING **STARCH** FROM TUBERS WHICH HAVE **STARCH** BRANCHING ENZYME CDNA SEQUENCE OPERABLY LINKED IN ANTISENSE ORIENTATION TO PROMOTER; HAS ELEVATED PEAK TEMPERATURE OF GELATINIZATION AND ELEVATED VICOSITY ONSET TEMPERATURE

IN Cooke David (GB); Gidley Michael John (GB); Jobling Stephen Alan (GB); Safford Richard (GB); Sidebottom Christopher Michael (GB); Westcott Roger J (GB)

PA National Starch and Chemical Investment Holding Corp (23275)

PI US 6103893 20000815
WO 9526407 19951005

AI US 1996-716449 19960924
WO 1995-GB634 19950322
19960924 PCT 371 date
19960924 PCT 102(e) date

PRAI GB 1994-6022 19940325
EP 1994-305806 19940804
EP 1995-300210 19950113

FI US 6103893 20000815

DT UTILITY

FS CHEMICAL

MRN 008908 MFN: 0595
009039 0583

NCL NCLM: 536102000
NCLS: 536128000

IC ICM: C08B030-00
ICS: C08B030-04; C08B030-20

TI HIGH AMYLOSE **STARCH** FROM TRANSGENIC **POTATO** PLANTS; EXTRACTING **STARCH** FROM TUBERS WHICH HAVE **STARCH**

BRANCHING ENZYME CDNA SEQUENCE OPERABLY LINKED IN ANTISENSE ORIENTATION TO PROMOTER; HAS ELEVATED PEAK TEMPERATURE OF GELATINIZATION AND ELEVATED VICOSITY. . .

AB Disclosed is a method of producing altered **starch** from transformed **potato** plants or their progeny, comprising extracting **starch** from a **potato** plant, at least the tubers of which comprise at least an effective portion of a **starch** branching enzyme (SBE) cDNA sequence operably linked in the antisense orientation to a suitable promoter, such that the level of SBE activity is limited to less than 0.8 units per gram tuber. Also disclosed are **potato** plants comprising altered **starch** in accordance with the invention.

ECLM D R A W I N G

1. A method of producing altered **starch** from transformed **potato** plants or their progeny, the method comprising extracting **starch** from a **potato** plant, at least the tubers of which comprise at least an effective portion of a **starch** branching enzyme (SBE) cDNA sequence operably linked in the antisense orientation to a suitable promoter, such that the level of. . .

ACLM 3. The method according to claim 1, wherein the **starch** is extracted from plants, the tubers thereof having less than 10% of the SBE activity in equivalent non-transformed plants.

4. The method according to claim 1, wherein the peak temperature of gelantinisation of the **starch** so produced is elevated by at least 2 degree(s) C. compared to unaltered **starch** produced from equivalent non-transformed plants.

5. The method according to claim 1, wherein the **viscosity onset** temperature of the **starch** so produced is elevated by at least 3 degree(s) C. compared to unaltered **starch** produced from equivalent non-transformed plants.

. . . The method according to claim 1, wherein the peak temperature of gelatinisation (as determined by differential scanning calorimetry) of the **starch** so produced is at least 71 degree(s) C.

7. The method according to claim 1, wherein the **viscosity onset** temperature of the **starch** so produced is at least 71 degree(s) C.

8. The method according to claim 1, comprising wet milling of **potato** tubers.

9. An altered **starch** produced by the method of claim 1.

10. The method according to claim 1, wherein the **starch** is extracted from plants, the tubers thereof having less than 5% of the SBE activity in equivalent non-transformed plants.

11. The method according to claim 1, wherein the peak temperature of gelatinization of the **starch** so produced is elevated by at least 5 degree(s) C. compared to unaltered **starch** produced from equivalent non-transformed plants.

12. The method according to claim 1, wherein the **viscosity onset** temperature of the **starch** so produced is elevated by at least 5 degree(s) C. compared to unaltered **starch** produced from equivalent non-transformed plants.

13. An altered **starch**, wherein the **starch** is extracted from transformed **potato** plants or their progeny, the plants having less than 0.8 units SBE activity per gram tuber, and, as extracted, the **starch** has the following physical properties: a) elevated peak temperature of gelatinisation as determined by differential scanning calorimetry (DSC) relative to unaltered **starch** extracted from equivalent non-transformed plants; and b) elevated **viscosity onset** temperature, relative to unaltered **starch** extracted from equivalent non-transformed plants.

14. The altered **starch** according to claim 13, wherein said **starch** is extracted from plants, the tubers thereof having less

than 10% SBE activity compared to equivalent non-transformed plants.
15. The altered **starch** according to claim 13, wherein the peak temperature of gelatinisation is elevated by at least 2 degree(s) C. compared to unaltered **starch** extracted from equivalent non-transformed plants.
16. The altered **starch** according to claim 13, wherein the **viscosity onset** temperature is elevated by at least 3 degree(s) C. compared to unaltered **starch** extracted from equivalent non-transformed plants.
17. The altered **starch** according to claim 13, wherein said **starch** is extracted from plants, the tubers thereof having less than 5% SBE activity compared to equivalent non-transformed plants.
18. The altered **starch** according to claim 13, wherein the peak temperature of gelatinization is elevated by at least 5 degree(s) C. compared to unaltered **starch** extracted from equivalent non-transformed plants.
19. The altered **starch** according to claim 13, wherein the **viscosity onset** temperature is elevated by at least 5 degree(s) C. compared to unaltered **starch** extracted from equivalent non-transformed plants.

L3 ANSWER 5 OF 12 PASCAL COPYRIGHT 2002 INIST-CNRS. ALL RIGHTS RESERVED.
TIEN Consequences of antisense RNA inhibition of **starch** branching enzyme activity on properties of **potato starch**
TIEN Consequences of antisense RNA inhibition of **starch** branching enzyme activity on properties of **potato starch**
AB Antisense constructs containing cDNAs for **potato starch** branching enzyme (SBE) were introduced into **potato** (Solanum tuberosum L.). A population of transgenic plants were generated in which tuber SBE activity was reduced by between 5. . . showed elevated phosphorous content. .sup.3.sup.1P n.m.r. analysis showed that this was due to proportionate increases in both 3- and 6-linked **starch** phosphates. A consistent alteration in **starch** gelatinisation properties was only observed when the level of SBE activity was reduced to below .eqvsim. 5% of that of. . . control values. Starches from these low SBE activity plants showed increases of up to 5.degree.C in d.s.c. peak temperature and **viscosity onset** temperature. Studies on melting of crystallites obtained from linear (1 4)-.alpha.-D-glucan oligomers suggest that an average difference of double helix. . .
CT 1,4-.alpha.-Glucan branching enzyme; Enzymatic activity; Inhibition; Transgenic plant; Antisense DNA; **Starch**; Biochemical analysis; Phosphates; Physicochemical properties; Solanum tuberosum; Gelatinization; Amylopectin; Amylose; Molecular structure; Carbohydrate; Metabolism

No Source

L3 ANSWER 6 OF 12 SCISEARCH COPYRIGHT 2002 ISI (R)
TI Consequences of antisense RNA inhibition of **starch** branching enzyme activity on properties of **potato starch**
TI Consequences of antisense RNA inhibition of **starch** branching enzyme activity on properties of **potato starch**
AB Antisense constructs containing cDNAs for **potato starch** branching enzyme (SBE) were introduced into **potato** (Solanum tuberosum L.). A population of transgenic plants were generated in which tuber SEE activity was reduced by between 5. . . showed elevated phosphorous P-31 n.m.r. analysis showed that this was due to proportionate increases in both content. 3- and 6-linked **starch** phosphates. A consistent alteration in **starch** gelatinisation properties war; only observed when the level of SEE activity was reduced to below similar to 5% of that. . . Starches from these low SEE activity plants showed increases of up to 5 degrees C in d.s.c. peak temperature and **viscosity onset** temperature. Studies

No Source

on melting of crystallites obtained from linear (1 --> 4)-alpha-D-glucan oligomers suggest that all average difference of double. . .

L3 ANSWER 7 OF 12 USPATFULL
TI High amylose **starch** from transgenic **potato** plants
AN 2000:106076 USPATFULL
TI High amylose **starch** from transgenic **potato** plants
IN Cooke, David, Oakley, United Kingdom
Gidley, Michael John, Raunds, United Kingdom
Jobling, Stephen Alan, Huntingdon, United Kingdom
Safford, Richard, Bedford, United Kingdom
Sidebottom, Christopher Michael, Bedford, United Kingdom
Westcott, Roger J., Wellingborough, United Kingdom
PA National Starch and Chemical Investment Holding Corporation, Wilmington, DE, United States (U.S. corporation)
PI US 6103893 20000815
WO 9526407 19951005
AI US 1996-716449 19960924 (8)
WO 1995-GB634 19950322
19960924 PCT 371 date
19960924 PCT 102(e) date
PRAI GB 1994-6022 19940325
EP 1994-305806 19940804
EP 1995-300210 19950113
DT Utility
FS Granted
LN.CNT 753
INCL INCLM: 536/102.000
INCLS: 536/128.000
NCL NCLM: 536/102.000
NCLS: 536/128.000
IC [7]
ICM: C08B030-00
ICS: C08B030-04; C08B030-20
EXF 435/91.1; 435/91.3; 435/93; 435/193; 435/101; 435/172.1; 435/172.3; 435/410; 435/417; 435/429; 435/320.1; 435/98; 435/210; 435/468; 435/469; 435/470; 514/44; 436/20; 536/23.2; 536/24.5; 536/102; 536/128; 800/205; 800/284; 800/286; 935/35
CAS INDEXING IS AVAILABLE FOR THIS PATENT.
TI High amylose **starch** from transgenic **potato** plants
AB Disclosed is a method of producing altered **starch** from transformed **potato** plants or their progeny, comprising extracting **starch** from a **potato** plant, at least the tubers of which comprise at least an effective portion of a **starch** branching enzyme (SBE) cDNA sequence operably linked in the antisense orientation to a suitable promoter, such that the level of SBE activity is limited to less than 0.8 units per gram tuber. Also disclosed are **potato** plants comprising altered **starch** in accordance with the invention.
SUMM This invention relates to a method of obtaining novel types of **starch** from **potato** plants, to novel **potato** plants from which the **starch** may be obtained, and to vectors for obtaining said plants.
SUMM **Starch** is the major form of carbon reserve in plants. constituting 50% or more of the dry weight of many storage organs--e.g. tubers. seeds of cereals. **Starch** is used in numerous food and industrial applications. In many cases, however. it is necessary to modify the native starches,. . . thereby removing the need for additional modification. To achieve this via genetic engineering requires knowledge of the metabolic pathway of **starch** biosynthesis. This includes characterisation of genes and encoded gene products which catalvse the synthesis of **starch**. Knowledge

about the regulation of **starch** biosynthesis raises the possibility of re-programming biosynthetic pathways to create starches with novel properties that could have new commercial applications.

SUMM The commercially useful properties of **starch** derive from the ability of the native granular form to swell and absorb water upon suitable treatment. Usually heat is. . . al., Cereal Foods World 33, 306-311. 1988) as " . . . the collapse (disruption) of molecular orders within the **starch** granule manifested in irreversible changes in properties such as granular swelling, native crvstallite melting, loss of birefringence. and **starch** solubilisation. The point of initial gelatinisation and the range over which it occurs is governed by **starch** concentration, method of observation, granule type, and heterogeneities within the granule population under observation". A number of techniques are available. . .

SUMM The consequence of the collapse of molecular orders within **starch** granules is that the granules are capable of taking up water in a process known as pasting, which has been. . . et al., Cereal Foods World 33, 306-311, 1988) as ". . . the phenomenon following gelatinisation in the dissolution of **starch**. It involves granular swelling, exudation of molecular components from the granule. and eventually. total disruption of the granules". The best. . . evaluating pasting properties is considered to be the viscoamylograph (Atwell et al., 1988) in which the viscosity of a stirred **starch** suspension is monitored under a defined time/temperature regime. A typical viscoamylograph profile for **potato starch** is shown in FIG. 5, in which the initial rise in viscosity is considered to be due to granule swelling.. . . observed due to increased fragmentation of swollen granules. This general profile (FIG. 5) has previously always been found for native **potato starch** . In addition to the overall shape of the viscosity response in a viscoamylograph, a convenient quantitative measure is the temperature of initial viscosity development (onset). FIG. 2 shows a typical viscosity profile for **starch** (Kennedy & Cabalda, Chem. in Britain. November 1991, 1017-1019), during and after cooking, with a representation of the physical state of the **starch** granules at various points. The letters A, B, C and D correspond to the stages of **viscosity onset** (A), maximum viscosity (B), complete dispersion (C) and re-association of molecules (or retrogradation, D).

SUMM The properties of **potato starch** are useful in a variety of both food and non-food (paper, textiles, adhesives etc.) applications. However, for many applications, properties. . . fragmentation during pasting as illustrated in FIG. 1. Currently the only ways of manipulating the gelatinisation and pasting temperatures of **potato starch** are by the inclusion of additives such as sugars, polyhydroxy compounds of salts (Evans and Haisman, Starke 34, 224-231, 1982). . . 1992) or by chemical cross-linking. Such processes are inconvenient and inefficient. It is therefore desirable to obtain plants which produce **starch** which intrinsically possesses such advantageous properties.

SUMM **Starch** Biosynthesis

SUMM **Starch** consists of 2 major components: amylose, a linear polymer of alpha, 1-4 linked glucose units; and amylopectin, a branched polvmer consisting of an alpha, 1-4 linked glucan backbone with alpha, 1-6 linked branches. The key enzymes in **starch** biosynthesis are the **starch** synthases and **starch** branching enzyme [alpha-1,4-glucan: alpha-1,4-glucan 6-glucosyltransferase. EC 2.4.1.18]. Amylose is synthesized from adenosine 5'-(alpha-D-glucopyranosyl pyrophosphate), or "ADP-glucose", by a **starch** synthase which is associated with the **starch** granule: the so-called "granule bound **starch** synthase" (GBSS). Amylopectin is synthesized from ADP-glucose by the concerted action of a soluble **starch** synthase (SSS) and **starch** branching enzyme (SBE). SBE

hydrolyses the linear alpha-1-4 glucan chain and rejoins the cleaved portion via an alpha-1-6 linkage to. . . a branched structure. The activity of SBE is thus of crucial importance in determining the type, and hence properties, of **starch** synthesized within plant systems.

SUMM **Starch** Branching Enzyme

SUMM . . . 24, 15-18 (1985); rice endosperm, Smyth. Plant Sci. 57, 1-8 (1988); pea embryo, Smith, Planta 175, 270-279 (1988)). However, in **potato** tuber, only a single form of SBE has so far been identified (Blennow & Johansson, Phytochem. 30, 437-444 (1991)).

SUMM . . . loss of SBE IIb. This reduction in SBE activity results in a higher ratio of amylose to amylopectin in endosperm **starch** compared to normal maize (Boyer & Preiss, Biochem. Biophys. Res. Commun. 80, 169-175 (1978)).

SUMM In pea embryos, 2 forms of SBE exist. The r (wrinkled) mutant of pea lacks SBE I activity and **starch** from this source has a higher ratio of amylose to amylopectin than normal peas [Smith, Planta 175, 270-279 (1988)].

SUMM In **potato**, amylose-free mutants have been obtained by X-ray irradiation (Hoverkamp-Hermelink et al., Theor. Appl. Genet. 75, 217-221, 1987) and by transformation with antisense-GBSS constructs (Visser et al., Mol. Gen. Genet. 225, 289-296, 1991). However, no high amylose mutants of **potato** exist and efforts to produce such via transformation with antisense SBE constructs have, hitherto, been unsuccessful (e.g. DE 41 04782A1).. . . antisense SBE technology, produced tubers containing only 10-20% SBE activity of control tubers, but: "neither the amylose content of the **starch** in the tubers of these plants, nor the total **starch** content of the tubers, was altered" (p.39). Similarly, WO 92/11375 suggests the use of an anti-sense approach to alter the **starch** content of tubers, but there was no reduction to practice and no data showing success of the approach, which disclosure. . .

SUMM . . . with even lower levels of SBE activity than those described by Wilmitzer. Surprisingly, especially in view of Wilmitzer's results, the **starch** obtained from such plants has unexpected novel, commercially useful properties.

SUMM In a first aspect the invention provides a method of producing altered **starch** from transformed **potato** plants or their progeny, the method comprising extracting **starch** from a **potato** plant, at least the tubers of which comprise at least an effective portion of a **starch** branching enzyme (SBE) cDNA sequence operably linked in the antisense orientation to a suitable promoter, such that the level of. . .

SUMM Altered **starch** produced according to the method of the invention is found to have the following physical properties:

SUMM a) elevated peak temperature of gelatinisation as determined by differential scanning calorimetry (DSC) relative to unaltered **starch** produced from equivalent non-transformed plants; and

SUMM b) elevated **viscosity onset** temperature, relative to unaltered **starch** produced from equivalent non-transformed plants.

SUMM The altered **starch** possesses these qualities ab initio as first extracted from the **potato** plant: the properties are not, for example, acquired by heating in the extraction process.

SUMM In a further aspect, the invention thus provides altered **starch** extracted from transformed **potato** plants or their progeny having less than 0.8 units SBE activity per gram tuber, the altered **starch** as extracted preferably having ab initio the properties defined above.

SUMM The parameters given above are frequently used by those skilled in the art to determine the properties of **starch**. The Examples below describe particular assay methods by which these parameters may be

determined.

SUMM . . . is the temperature at which there is a maximum in the loss of order in granules within a sample of **starch** in the presence of excess water, as judged by the heat flow required to maintain a constant rate of temperature. . . temperature of gelatinisation is elevated by at least 2.degree. C. more preferably by at least 5.degree. C., compared to unaltered **starch**.

SUMM For the purposes of the present specification, the **viscosity onset** temperature is defined as the temperature at which the viscosity of a 10% w/w aqueous **starch** solution becomes at least 50% greater than the maximum viscosity of the solution at lower temperatures (above 50.degree. C.). Viscosity may be measured in arbitrary units (e.g. instrument stirring number units or "SNU"). Preferably the **viscosity onset** temperature is elevated by at least 3.degree. C., and more preferably by at least 5.degree., C. compared to unaltered **starch**.

SUMM Preferably the altered **starch** produced from the transformed plants (or the progeny thereof) has a peak temperature of gelatinisation (as determined by differential scanning caloriometry) of at least 71.degree. C. and/or a **viscosity onset** temperature of at least 71.degree. C.

SUMM The altered **starch** is extracted from **potato** plants in which the **starch** branching enzyme (SBE) activity is less than 0.8 units per gram tuber. (A unit of activity is defined for present purposes as the amount of enzyme activity which incorporates into **starch** 1 micromole of glucose per minute at a temperature of 30.degree. C.)

SUMM Preferably the altered **starch** is extracted from the plant by wet milling of **potato** tubers.

SUMM Preferably the altered **starch** is obtained from transformed **potato** plants or their progeny, the tubers of which exhibit less than 10%, and preferably less than 5%, of SBE activity. . .

SUMM In a further aspect, the invention provides a vector for modifying a **potato** plant so as to cause the plant to be capable of giving rise to tubers having less than 0.8 units. . .

SUMM Preferably the vector comprises a full length SBE CDNA sequence, preferably that of **potato** SBE, operably linked in the antisense orientation to a suitable promoter. Suitable promoters include the CaMV 35S and the GBSS. . .

SUMM In another aspect the invention provides a **potato** plant capable of giving rise to tubers having less than 0.8 units SBE activity per gram tuber and comprising at. . .

DRWD FIG. 1 shows how the degree of gelatinisation of an unaltered **starch** sample varies with temperature, as measured by differential scanning calorimetry;

DRWD FIG. 2 shows the typical viscosity profile of conventional **starch** during and after cooking, together with representations of the physical state of **starch** granules at various stages;

DRWD FIG. 3 shows how the degree of gelatinisation of a sample of altered **starch** in accordance with the invention varies with temperature as measured by differential scanning calorimetry (DSC);

DRWD FIG. 5 is a graph of viscosity (SNU) against temperature (.degree. C.) for unaltered **starch**;

DRWD FIG. 6 is a graph of **viscosity onset** temperature (.degree. C.) against SBE activity (Units), showing how the two parameters are related;

DRWD FIG. 7 is a graph of viscosity (SNU) against temperature (.degree. C.) for altered **starch** in accordance with the invention; and

DRWD FIG. 8 shows the sequence, SEQ ID NO. 1, of a full length **potato** SBE cDNA clone.

DETD Construction of Plant Transformation Vectors containing Antisense **Starch** Branching Enzyme Genes

DETD (a) Construction of Enhanced 35S Antisense **Potato** **Starch** Branching Enzyme Plant Transformation Vector

DETD Initally a 1.4 kb EcoRI partial length cDNA for **potato** **starch** branching enzyme was purchased from the Agricultural Genetics Company (Cambridge, UK). This cDNA was isolated from a lambda phage library (methylase protected fragments) made from RNA extracted from **potato** tubers (cv Desiree) using standard techniques (Sambrook. Fritsch & Maniatis, (1989) Molecular Cloning: A Laboratory Manual, Cold Spring Harbor Lab,. . .

DETD . . . conducted. The full length sequence shown in FIG. 8 is in reasonably close agreement with the full length sequence of **potato** SBE disclosed by Poulsen & Kreiberg (1993, Plant Physiol. 102, 1053-1054), although some differences are readily apparent. Other SBE sequences. . .

DETD . . . (1980) Cell 21, 285-294) in the vector pJIT 60 (Guerineau et al., (1992) Plant Mol. Biol: 18, 815-818). The promoter-antisense **potato** **starch** branching enzyme-polyA fragment was then cloned into the plant transformation vector BIN19 (Bevan M (1984) Nucl. Acids Res. 12, 8711-8721).

DETD (b) Construction of Patatin Promoter Antisense **Potato** **Starch** Branching Enzyme Plant Transformation Vector

DETD The 2.3 kb EcoRI fragment (corresponding to about 2/3 of the full length cDNA) of the **potato** **starch** branching enzyme was subcloned into the EcoRI site of the pBSSK II plus vector (Stratagene) to create pSJ5. A Xhol. . .

DETD Transformation of **Potato** with Antisense **Starch** Branching Enzyme Constructs

DETD Stock nodal cutting cultures of **potato** (cv. Desiree) were maintained on Murashige and Skoog basal media (MS) containing 1% sucrose at 22.degree. C. in an illuminated. . .

DETD . . . transferred to Genescreen in 20.times.SSC and u.v. cross-linked (Stratalinker, Stratgene). Blots were hybridised to random-prime labelled (Amersham) 2.3 kb EcoRI **potato** **starch** branching enzyme fragment in 5.times.SSPE (0.9M NaCl, 50 mM NaH.sub.2 PO.sub.4, 5 mM EDTA), 5.times.Denhardts solution, 1% SDS, 100 .mu.g/ml. . .

DETD (b) **Starch** Branching Enzyme (SBE) Assay of Transgenic Tubers

DETD . . . crude homogenate was clarified by centrifuging at 10.000 g for 10 minutes. The supernatant was retained for the assay of **starch** branching enzyme activity.

DETD . . . mM 2-(N-morpholino) ethanesulphonic acid (MES) buffer, pH 6.5, 50 mM[.sup.14 C]glucose 1-phosphate (100 nCi), 0.05 mg rabbit phosphorylase A and **potato** tuber extract. Incubations were performed at 30.degree. C. for 60 minutes. Negative controls contained either: (a) no phosphorylase, or (b) the **potato** tuber extract boiled for 30 minutes to destroy enzyme activity. The reaction was terminated and glucan polymer precipitated by the. . .

DETD **Starch** Branching Enzyme Assays of Transgenic **Potato** Tuber Extracts

DETD All **starch** branching enzyme activities were measured in duplicate and mean values taken. At low levels of activity absolute quantitation, via the. . .

DETD

POTATO TUBER **STARCH** BRANCHING ENZYME ACTIVITY		
PLANT (units g.sup.-1 tuber)		ACTIVITY
CONTROL	58	21.3
40	18.2	
31	16.6	
29	13.1	
49	13.0	

8 12.7

. . .

DETD Analysis of Transgenic **Starch** Properties

DETD (a) **Starch** Extraction

DETD **Potato** tubers were homogenised in water for 2 min in a Waring blender operating at high speed. The homogenate was washed. . . (initially 2 mm, then 1 mm filters) using approximately 4L of water per 100 g of tubers (6 extractions). Washed **starch** granules were finally extracted with acetone and air dried.

DETD . . . temperature range for the loss of granule order upon heating starches in excess water was determined by differential scanning calorimetry. **Starch** powders isolated from a range of transgenic **potato** plants were analyses using the Perkin Elmer DSC 7 instrument. 1-4 mg of **starch** was accurately weighed into an aluminium sample pan, and water added so that the **starch** concentration was less than 25% w/v, to give a total sample weight of 10-15 mg. An empty reference sample pan. . .

DETD Starches isolated from **potato** plants exhibiting a range of **starch** branching enzyme activities (determined as described in Example 3b) were characterised by differential scanning calorimetry. Peak temperatures are compared with **starch** branching enzyme activity in FIG. 4. from which it appears that levels of enzyme activity less than 0.8 U/g of. . .

DETD Starches isolated from a range of transgenic **potato** plants were analysed for viscosity development (`pasting`) following the loss of granule order. The instrument used was the Rapid Visco Analyser 3C (Newport Scientific, Sydney, Australia). **Starch** (2.50 g) was weighed into an instrument sample holder, and water (22.50 g) added so that the final concentration was 10% w/w **starch**. Suspensions were equilibrated for 2 minutes at 50.degree. C. and heated under standard stirring conditions at 1.5.degree. C. minute from. . . but the fact that viscosity starts from a very low level and rapidly rises allows an accurate determination of a **viscosity onset** temperature, defined as the temperature at which viscosity is at least 50% higher than at all lower temperatures above 50.degree.. . .

DETD The **viscosity onset** temperatures for starches isolated from **potato** plants exhibiting a range ot **starch** branching enzyme activities were determined, with the results shown in FIG. 6. These data show that a consistent increase in **viscosity onset** temperature is found for starches from plants containing less than 0.8 U/g of tuber of **starch** branching enzyme. For those starches which show a higher **viscosity onset** temperature, other parameters of pasting (e.g. peak temperature) are also higher. This is illustrated by comparison of FIGS. 5 (onset. . .

DETD Construction of GBSS antisense full lenoth **potato** **starch** branching enzyme vector

DETD The inventors have recently made a further construct comprising a full length **potato** SBE cDNA in the anti-sense orientation under the control of the GBSS promoter. Details of the construction are given below.. . .

DETD A full length cDNA clone for **potato** **starch** branching enzyme corresponding to nucleotides 91-3114 plus an additional 10 bases at the 3' end (Poulsen, P. & Kreiberg, J. D. Plant Physiol. (1993) 102: 1053-1054) was isolated from a **potato** tuber cDNA library (see above). The cDNA was excised from the plasmid vector by cutting with SacI and XhoI and inserted in an antisense orientation between the granule bound **starch** synthase promoter (GBSS) and the nos polyadenylation signal in the BIN 19 based plant transformation vector pPGB121 which had been cut with SacI and SalI. The GBSS promoter is a 0.8 kb HindIII-NsiI fragment of the granule bound **starch** synthase genomic clone LGBSSwt-6; this promoter fragment directs GUS

expression in an organ specific manner (up to 3350 fold higher. . .
DETD . . . TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
- - (ii) MOLECULE TYPE:
(A) DESCRIPTION: cDNA e - #ncoding **starch** branching enzyme
- - (iii) HYPOTHETICAL: No
- - (iv) ANTI-SENSE: No
- - (v) FRAGMENT TYPE:
- - (vi). . . (viii) POSITION IN GENOME:
(A) CHROMOSOME/SEGMENT:
(B) MAP POSITION:
(C) UNITS:
- - (ix) FEATURE: open reading frame
(A) NAME/KEY: **starch** br - #anching enzyme
(B) LOCATION: 44-2788
(C) IDENTIFICATION METHOD: - # lone ORF with homology to other **starch**
branching - #enzymes
(D) OTHER INFORMATION: - #complements KV832 E. coli glycogen
branching - #enzyme mutant
- - (xi) SEQUENCE. . .

CLM What is claimed is:

1. A method of producing altered **starch** from transformed **potato** plants or their progeny, the method comprising extracting **starch** from a **potato** plant, at least the tubers of which comprise at least an effective portion of a **starch** branching enzyme (SBE) cDNA sequence operably linked in the antisense orientation to a suitable promoter, such that the level of. . .

3. The method according to claim 1, wherein the **starch** is extracted from plants, the tubers thereof having less than 10% of the SBE activity in equivalent non-transformed plants.

4. The method according to claim 1, wherein the peak temperature of gelantinisation of the **starch** so produced is elevated by at least 2.degree. C. compared to unaltered **starch** produced from equivalent non-transformed plants.

5. The method according to claim 1, wherein the **viscosity onset** temperature of the **starch** so produced is elevated by at least 3.degree. C. compared to unaltered **starch** produced from equivalent non-transformed plants.

. . . The method according to claim 1, wherein the peak temperature of gelatinisation (as determined by differential scanning calorimetry) of the **starch** so produced is at least 71.degree. C.

7. The method according to claim 1, wherein the **viscosity onset** temperature of the **starch** so produced is at least 71.degree. C.

8. The method according to claim 1, comprising wet milling of **potato** tubers.

9. An altered **starch** produced by the method of claim 1.

10. The method according to claim 1, wherein the **starch** is extracted from plants, the tubers thereof having less than 5% of the SBE activity in equivalent non-transformed plants.

11. The method according to claim 1, wherein the peak temperature of

gelatinization of the **starch** so produced is elevated by at least 5.degree. C. compared to unaltered **starch** produced from equivalent non-transformed plants.

12. The method according to claim 1, wherein the **viscosity onset** temperature of the **starch** so produced is elevated by at least 5.degree. C. compared to unaltered **starch** produced from equivalent non-transformed plants.

13. An altered **starch**, wherein the **starch** is extracted from transformed **potato** plants or their progeny, the plants having less than 0.8 units SBE activity per gram tuber, and, as extracted, the **starch** has the following physical properties: a) elevated peak temperature of gelatinisation as determined by differential scanning calorimetry (DSC) relative to unaltered **starch** extracted from equivalent non-transformed plants; and b) elevated **viscosity onset** temperature, relative to unaltered **starch** extracted from equivalent non-transformed plants.

14. The altered **starch** according to claim 13, wherein said **starch** is extracted from plants, the tubers thereof having less than 10% SBE activity compared to equivalent non-transformed plants.

15. The altered **starch** according to claim 13, wherein the peak temperature of gelatinisation is elevated by at least 2.degree. C. compared to unaltered **starch** extracted from equivalent non-transformed plants.

16. The altered **starch** according to claim 13, wherein the **viscosity onset** temperature is elevated by at least 3.degree. C. compared to unaltered **starch** extracted from equivalent non-transformed plants.

17. The altered **starch** according to claim 13, wherein said **starch** is extracted from plants, the tubers thereof having less than 5% SBE activity compared to equivalent non-transformed plants.

18. The altered **starch** according to claim 13, wherein the peak temperature of gelatinization is elevated by at least 5.degree. C. compared to unaltered **starch** extracted from equivalent non-transformed plants.

19. The altered **starch** according to claim 13, wherein the **viscosity onset** temperature is elevated by at least 5.degree. C. compared to unaltered **starch** extracted from equivalent non-transformed plants.

L3 ANSWER 8 OF 12 WPIDS (C) 2002 THOMSON DERWENT
TI **Potato starch** stable against freeze-thaw cycles in native form, useful e.g. as thickener for foods, produced from transgenic plants ~~in~~ which **starch** synthase enzymes are inhibited.
AN 2001-257879 [26] WPIDS
DNC ~~C2001-077719~~
TI **Potato starch** stable against freeze-thaw cycles in native form, useful e.g. as thickener for foods, produced from transgenic plants in which **starch** synthase enzymes are inhibited.
DC C06 D16 D17
IN JOBLING, S A; SCHWALL, G P; WESTCOTT, R J
PA (NATT) NAT STARCH & CHEM INVESTMENT HOLDING COR
CYC 95
PI WO 2001019975 A2 20010322 (200126)* EN 76p C12N015-00

RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ
NL OA PT SD SE SL SZ TZ UG ZW
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CR CU CZ DE DK DM
DZ EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC
LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ PL PT RO RU SD SE
SG SI SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW

AU 2000070310 A 20010417 (200140) C12N015-00

EP 1212440 A2 20020612 (200239) EN C12N015-82

R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT
RO SE SI

ADT WO 2001019975 A2 WO 2000-GB3522 20000913; AU 2000070310 A AU 2000-70310 20000913; EP 1212440 A2 EP 2000-958901 20000913, WO 2000-GB3522 20000913

FDT AU 2000070310 A Based on WO 200119975; EP 1212440 A2 Based on WO 200119975

PRAI GB 1999-21830 19990915

IC ICM C12N015-00; C12N015-82

ICS A01H005-00; C08B030-00; C08B030-12; C12N009-10

TI **Potato starch** stable against freeze-thaw cycles in native form, useful e.g. as thickener for foods, produced from transgenic plants in which **starch** synthase enzymes are inhibited.

AB WO 200119975 UPAB: 20010515

NOVELTY - **Potato starch** (I), in native form as extracted, has freeze-thaw stability such that a 1 %weight/volume aqueous suspension has an absorbance at. . . INDEPENDENT CLAIMS are also included for the following:

(a) plant cell containing nucleic acid sequences (I) that specifically inhibit granule-bound **starch** synthase I (GBSSI) and at least one other enzyme involved in **starch** synthesis;

(b) plant cell containing (I) that specifically inhibit expression of 3 or more enzymes involved in **starch** synthesis;

(c) plants, or their progeny, containing (I), that specifically inhibit granule-bound **starch** synthase I (GBSSI) and at least one other enzyme involved in **starch** synthesis;

(d) method (M1) of altering the **starch** composition of plants by introducing (I) that specifically inhibit granule-bound **starch** synthase I (GBSSI) and at least one other enzyme involved in **starch** synthesis;

(e) plants produced by (M1);

(f) **starch** produced by plants (produced by M1); and

(g) generally any **potato starch** that, in native form as extracted, has freeze-thaw stability.

USE - (I) is used in preparation of thickener compositions. . .

TECH UPTX: 20010515

TECHNOLOGY FOCUS - BIOLOGY - Preferred **starch**: (I) is also charcterized by:

(1) freeze-thaw stability such that a 5 %weight/volume (% w/v) aqueous paste shows less than 40. . . than 8%, and ratio of fraction I:fraction II short chain glucans of at least 60, preferably 70,%; optionally also a **viscosity onset** temperature below 67 degrees Celsius (best 51 degrees Celsius) (as determined by viscometric analysis of a 7.4% w/v aqueous suspension,. . . DSC (differential scanning calorimter) 7, using a 10 mg sample in an aqueous mixture of less than 25 % w/v **starch** content. After extraction, (I) may be modified by physical, chemical and/or enzymatic processes.

TECHNOLOGY FOCUS - BIOTECHNOLOGY - Preferred plants: In the cells, at least one of **starch** synthase (SS) II and/or III is inhibited, particularly by expressing the appropriate antisense sequence or by sense suppression methods. The transformed cells are regenerated to plants conventionally. The cells are particularly from **potato** but may also be from rice, cassava or maize.

TT TT: **POTATO STARCH** STABILISED FREEZE THAW CYCLE NATIVE FORM USEFUL THICKEN FOOD PRODUCE TRANSGENIC PLANT **STARCH**

SYNTHASE ENZYME INHIBIT.

L3 ANSWER 9 OF 12 WPIDS (C) 2002 THOMSON DERWENT
TI Altering characteristics of plants by inhibiting expression of **starch** synthase enzymes, used to produce modified **starch** with reduced **viscosity onset** temperature.
AN 2000-126546 [11] WPIDS
DNN N2000-095371 DNC C2000-038539
TI Altering characteristics of plants by inhibiting expression of **starch** synthase enzymes, used to produce modified **starch** with reduced **viscosity onset** temperature.
DC A11 A97 C06 D16 F06 F09 G03 P13
IN EDWARDS, E A; JOBLING, S A; MARTIN, C R; SCHWALL, G P; SMITH, A M; WESTCOTT, R J
PA (NATT) NAT STARCH & CHEM INVESTMENT HOLDING COR
CYC 87
PI WO 9966050 A1 19991223 (200011)* EN 49p C12N015-54
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL OA PT SD SE SL SZ UG ZW
W: AE AL AM AT AU AZ BA BB BG BR BY CA CH CN CU CZ DE DK EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT UA UG US UZ VN YU ZA ZW
AU 9943802 A 20000105 (200024) C12N015-54
EP 1092033 A1 20010418 (200123) EN C12N015-54
R: AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE
JP 2002518015 W 20020625 (200243) 55p C12N015-09
ADT WO 9966050 A1 WO 1999-GB1902 19990615; AU 9943802 A AU 1999-43802 19990615; EP 1092033 A1 EP 1999-926617 19990615, WO 1999-GB1902 19990615; JP 2002518015 W WO 1999-GB1902 19990615, JP 2000-554859 19990615
FDT AU 9943802 A Based on WO 9966050; EP 1092033 A1 Based on WO 9966050; JP 2002518015 W Based on WO 9966050
PRAI EP 1998-304716 19980615
IC ICM C12N015-09; C12N015-54
ICS A01H005-00; A21D002-36; C08B030-04; C12N015-82
TI Altering characteristics of plants by inhibiting expression of **starch** synthase enzymes, used to produce modified **starch** with reduced **viscosity onset** temperature.
AB . . .
least two sequences (I), or their fragments or functional equivalents, each comprising a gene that encodes an enzyme (II) with **starch** synthase activity. Each (I) is linked to a promoter and is able to affect expression of the corresponding endogenous gene.
. . . INDEPENDENT CLAIMS are also included for the following:
(a) plants modified this way and their progeny or parts;
(b) **starch** obtained from the plants of (a);
(c) **starch** obtained from **potato** and having
(i) **viscosity onset** temperature less than 55 deg. C (measured at atmospheric pressure on a 10wt.% aqueous suspension in a Newport Scientific Rapid. . . endosperm onset temperature less than 50 deg. C (measured by differential scanning calorimetry on a Perkin-Elmer DSC7);
(d) production of **starch** by extracting plants of (a);
(e) nucleic acid construct (A) containing at least two (I), operably linked to a promoter;. . . (f) plants, or their parts or progeny, containing (A).
ACTIVITY - None given.
MECHANISM OF ACTION - Altering activity of **starch** synthesis enzymes, by sense or antisense suppression of endogenous genes.
USE - The modified plants are used to produce **starch** for use in production or processing of foods, paper, textiles and adhesives (claimed).

ADVANTAGE - **Starch** produced by the transgenic plants has a lower **viscosity onset** temperature, so requires milder processing conditions (reduced energy demand) and, in foods, imparts better quality and color with reduced off-flavors. . .

TECH UPTX: 20000301

TECHNOLOGY FOCUS - BIOLOGY - Preferred **Starch**: This has (i) **viscosity onset** temperature (measured as above) at least 10, preferably 12,degreesC lower than that of **starch** from unmodified plants;
(ii) endosperm onset temperature (measured as above) at least 15, especially 17,degreesC lower (best less than 44degreesC) and. . . anion-exchange chromatography on a Dionex Carbopac DA-100 column). The specification includes a graph comparing DP distribution for modified and native **starch**.

TECHNOLOGY FOCUS - BIOTECHNOLOGY - Preferred Process: All (I) are introduced simultaneously, particularly on a single nucleic acid construct, or one. . . orientation with respect to the promoter (which may be constitutive or tissue-specific). Particularly (I) are derived from the genes for **starch** synthases (SS) II and (III), especially from **potato**, or their equivalents.
Preferred plants: These are **potato** (most preferred), cassava, maize, wheat, barley, tomato, rice and pea.
Preparation: Transfer vectors are made by standard methods, e.g. isolating. . .

TT TT: ALTER CHARACTERISTIC PLANT INHIBIT EXPRESS **STARCH** SYNTHASE ENZYME PRODUCE MODIFIED **STARCH** REDUCE **VISCOSITY ONSET** TEMPERATURE.

L3 ANSWER 10 OF 12 WPIDS (C) 2002 THOMSON DERWENT
TI New isolated **potato** isoamylase-type debranching enzyme gene.
AN 1999-229220 [19] WPIDS
DNC C1999-067421
TI New isolated **potato** isoamylase-type debranching enzyme gene.
DC C06 D16 D17
IN JOBLING, S A; SCHWALL, G P; WESTCOTT, R J
PA (NATT) NAT STARCH & CHEM INVESTMENT HOLDING COR
CYC 21
PI WO 9912950 A2 19990318 (199919)* EN 50p C07H021-00
RW: AT BE CH CY DE DK ES FI FR GB GR IE IT LU MC NL PT SE
W: AU CA US
AU 9889911 A 19990329 (199932) C07H021-00
EP 1009751 A2 20000621 (200033) EN C07H021-00
R: AT BE DE DK ES FI FR GB GR IT NL PT SE
ADT WO 9912950 A2 WO 1998-GB2665 19980904; AU 9889911 A AU 1998-89911 19980904; EP 1009751 A2 EP 1998-941593 19980904, WO 1998-GB2665 19980904
FDT AU 9889911 A Based on WO 9912950; EP 1009751 A2 Based on WO 9912950
PRAI GB 1997-18863 19970906
IC ICM C07H021-00
TI New isolated **potato** isoamylase-type debranching enzyme gene.
AB WO 9912950 UPAB: 19990518
NOVELTY - (A) A novel nucleic acid sequence is obtainable from **potato** plants and carries at least a portion of an isoamylase-type debranching enzyme (DBE) gene.
DETAILED DESCRIPTION - INDEPENDENT CLAIMS are. . . altered by a method as in (5) or the progeny.
USE - The constructs can be used to alter the **starch** properties of plants such as **potato**, sweet **potato**, maize, wheat, barley, oat, cassava, pea or rice (claimed). The **starch** can have increased branching and/or shorter chain length, reduced peak viscosity, higher setback viscosity or increased **viscosity onset** temperature (claimed).

ADVANTAGE - By using an antisense sequence with greater homology to the native gene, greater inhibition can be. . .

TECH. . .
and
(2) ATG GAT GTT GTH TWY AAY CAT (DBE1).
These were used in a PCR reaction using first strand cDNA from **potato** leaf or tuber RNA or genomic DNA as template. The product was used to obtain a 1.5kb DNA sequence. The. . . encoded by the sugary-1 gene. A PCR based cloning strategy was also used for isolating pullulanase type debranching enzymes from **potato**, using conserved domains within the known cloned gene sequences, of which spinach was the only plant gene (GenBank SOPULSPO1). A. . .

TT TT: NEW ISOLATE **POTATO** ISOAMYLASE TYPE ENZYME GENE.

L3 ANSWER 11 OF 12 WPIDS (C) 2002 THOMSON DERWENT
TI Soluble **starch** synthase - used to produce altered **starch** from commercially important plants, e.g. **potato**, rice, wheat, and maize.
AN 1997-312737 [29] WPIDS
DNN N1997-258924 DNC C1997-100802
TI Soluble **starch** synthase - used to produce altered **starch** from commercially important plants, e.g. **potato**, rice, wheat, and maize.
DC C06 D16 D17 P13
IN EDWARDS, E A; MARSHALL, J; MARTIN, C R; SMITH, A M
PA (NATT) NAT STARCH & CHEM INVESTMENT HOLDING COR
CYC 19
PI EP 779363 A2 19970618 (199729)* EN 39p C12N015-82
R: AT BE CH DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE
AU 9674268 A 19970703 (199735) C12N009-10
AU 723475 B 20000824 (200045) C12N009-10
ADT EP 779363 A2 EP 1996-309004 19961211; AU 9674268 A AU 1996-74268 19961211; AU 723475 B AU 1996-74268 19961211
FDT AU 723475 B Previous Publ. AU 9674268
PRAI GB 1995-25353 19951212
IC ICM C12N009-10; C12N015-82
ICS A01H005-00; C08B030-00; C12N005-10; C12N015-05; C12N015-54
TI Soluble **starch** synthase - used to produce altered **starch** from commercially important plants, e.g. **potato**, rice, wheat, and maize.
AB EP 779363 UPAB: 19970716
A novel altered **starch** (A), is extracted from a transformed **potato** plant or its progeny, and as extracted, has a **viscosity onset** temperature (VOT) as determined by differential scanning calorimetry (DSC) that is reduced by at least 5 deg. C compared to **starch** extracted from equivalent, non-transformed plants. Also new are: (1) a polypeptide obtained from a soluble extract of **potato** tubers, having **starch** synthase (SS) activity; (2) a nucleic acid sequence of at least 200 bp, which exhibits at least 80% sequence identity with the corresponding region of the 4127 bp DNA sequence of **potato** soluble SS (given in the specification), operably linked in the sense or antisense orientation to a promoter functional in a. . . plant of (5), or the sequences of (1), (2), or (3) can be used in a method to produce altered **starch** from a **potato** plant, or other commercially important plants, such as tomato, rice, wheat, peal cassava, sweet **potato**, barley, oat or maize (claimed).
ADVANTAGE - The soluble SS has a specific activity which is greater than other SSs. The **starch** produced by this enzyme has significantly altered physical properties (such as VOT).
Dwg.0/7
TT TT: SOLUBLE **STARCH** SYNTHASE PRODUCE ALTER **STARCH**

COMMERCIAL IMPORTANT PLANT **POTATO** RICE WHEAT MAIZE.

L3 ANSWER 12 OF 12 WPIDS (C) 2002 THOMSON DERWENT
TI New transformed **potato** plants or their progeny - contg. anti sense **starch** branching enzyme cDNA used for producing **starch** with altered properties.
AN 1995-351326 [45] WPIDS
DNN N1995-261962 DNC C1995-153906
TI New transformed **potato** plants or their progeny - contg. anti sense **starch** branching enzyme cDNA used for producing **starch** with altered properties.
DC C06 D16 D17 F06 F09 G03 P13
IN COOKE, D; GIDLEY, M J; JOBLING, S A; SAFFORD, R; SIDEBOTTOM, C M; WESTCOTT, R J
PA (NATT) NAT STARCH & CHEM INVESTMENT; (NATT) NAT STARCH & CHEM INVESTMENT HOLDING COR
CYC 58
PI WO 9526407 A1 19951005 (199545)* EN 36p C12N015-82
RW: AT BE CH DE DK ES FR GB GR IE IT KE LU MC MW NL OA PT SD SE SZ UG
W: AM AT AU BB BG BR BY CA CH CN CZ DE DK ES FI GB GE HU JP KE KG KP KR KZ LK LT LU LV MD MG MN MW NL NO NZ PL PT RO RU SD SE SI SK TJ TT UA US UZ VN
AU 9519028 A 19951017 (199604) C12N015-82
EP 754235 A1 19970122 (199709) EN C12N015-82
R: AT BE DE ES FR GB IT NL SE
AU 688006 B 19980305 (199820) C12N015-82
US 6103893 A 20000815 (200041) C08B030-00
CA 2186399 C 20010904 (200155) EN C12N015-54
ADT WO 9526407 A1 WO 1995-GB634 19950322; AU 9519028 A AU 1995-19028 19950322; EP 754235 A1 EP 1995-911460 19950322, WO 1995-GB634 19950322; AU 688006 B AU 1995-19028 19950322; US 6103893 A WO 1995-GB634 19950322, US 1996-716449 19960924; CA 2186399 C CA 1995-2186399 19950322, WO 1995-GB634 19950322
FDT AU 9519028 A Based on WO 9526407; EP 754235 A1 Based on WO 9526407; AU 688006 B Previous Publ. AU 9519028, Based on WO 9526407; US 6103893 A Based on WO 9526407; CA 2186399 C Based on WO 9526407
PRAI EP 1995-300210 19950113; GB 1994-6022 19940325; EP 1994-305806 19940804
IC ICM C08B030-00; C12N015-54; C12N015-82
ICS A01H005-00; C08B030-04; C08B030-14; C08B030-20; C12N015-11
TI New transformed **potato** plants or their progeny - contg. anti sense **starch** branching enzyme cDNA used for producing **starch** with altered properties.
AB WO 9526407 UPAB: 19951114
Transformed **potato** plants or their progeny capable of giving rise to tubers having altered **starch**, comprising at least an effective portion of a **starch** branching enzyme (SBE) cDNA operably linked in the antisense orientation to a suitable promoter, such that the level of SBE. . . to less than 0.8 units per gram of tuber, are claimed. Also claimed are: (1) a vector for modifying a **potato** plant so as to cause the plant to be capable of giving rise to tubers having less than 0.8 units. . . least an effective portion of a SBE cDNA operably linked in the antisense orientation to a suitable promoter; (2) altered **starch** extracted from transformed **potato** plants or their progeny, the plants having less than 0.8 units SBE activity per gram of tuber, where the extracted **starch** has the following physical properties: (i) an elevated peak temp. of gelatinisation (detd. by differential scanning calorimetry (DSC)), relative to unaltered **starch** extracted from equivalent non-transformed plants; and (ii) an elevated **viscosity onset** temp., relative to unaltered **starch** extracted from equivalent non-transformed plants.

USE - The starches can be used in food and non-food (e.g. paper, textiles. . . ADVANTAGE - The plants have low levels of SBE activity, and produce starches with elevated peak temps. of gelatinisation and **viscosity onset**.

Dwg.0/8

TT TT: NEW TRANSFORM **POTATO** PLANT PROGENY CONTAIN ANTI SENSE **STARCH** BRANCH ENZYME CDNA PRODUCE **STARCH** ALTER PROPERTIES.